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ORIGINAL ARTICLE

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Biological monitoring of carbon disulphide and phthalate exposure in the contemporary rubber industry

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Abstract *Objectives*: We studied the range in urinary levels of 2-thiothiazolidine-4-carboxyl acid (TTCA), a metabolite of CS_2 and phthalic acid (PA), a common metabolite of phthalates, across factories and departments in the contemporary rubber manufacturing industry. *Methods*: Spot urine samples from 101 rubber workers employed in nine different factories were collected on Sunday and during the workweek on Tuesday, Wednesday and Thursday at ~4 pm. In total, 386 urine samples were successfully analyzed. *Results*: Levels of both biomarkers increased significantly by a factor 2 (paired *t*-test *P*-value <0.05) during the working week as compared to the Sunday biomarker levels with absolute increases of approximately 70 μg/l and 5 μmol/mol creatinine for PA and TTCA, respectively. Levels in both biomarkers did not differ markedly between working days. Increases seemed to be restricted to specific factories and/or departments (e.g. molding and curing). *Conclusions*: The results of this study demonstrate that rubber workers in the contemporary rubber industry are exposed to phthalates and low levels of CS_2 (~0.05 ppm) as measured by PA and TTCA, respectively. Exposures to both compounds are largely driven by specific circumstances in factories. Therefore, when estimating exposures to phthalates and CS_2 detailed information should be collected on the type and amount of phthalate containing ester plasticizers, dithiocarbamates and thiurams used. Preferably, personal exposure data should be collected. In this case, biological monitoring seems a reasonable approach. However, in the case of PA attention should be given to individual background levels as this could lead to a substantial overestimation of the occupational contribution to total phthalate exposure.

Keywords Rubber industry · Carbon disulphide · Phthalates · Biomonitoring

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Introduction

Exposures in the rubber industry are multitudinous and have given rise to several occupational health concerns, among others, cancer, cardiovascular disease, pulmonary function abnormalities, hypertension, deterioration of intellectual and psychomotor function, nervous system dysfunction, and reproductive disorders (Roth 1999). Most of the epidemiological studies conducted so far have focused on the cancer risk in this industry. However, recently several contemporary cohort studies among rubber workers in the United Kingdom, Germany and Sweden have been initiated where in at least in one of these cohorts other health outcomes than cancer are being studied such as reproductive disorders, cardiovascular, and airways diseases (Mikoczy et al. 2004a, b). Historically, exposure to carbon disulphide (CS_2) has been linked to cardiovascular diseases within this industry (Danilenko and Kozintseva 1989; Delpech 1863; Roth 1999). Reproductive disorders have been observed in several studies among rubber workers and it has been hypothesized that this might be linked to exposure to rubber chemicals among, which CS_2 and/or phthalates (Duty et al. 2003; Figa-Talamanca 1984).

Phthalates are widely used in the rubber industry as ester plasticizers. The most commonly used phthalate compounds are di- *n*-butyl phthalate (DBP) and di-isooctyl phthalate (DIOP), but others like bis(2-ethylhexyl) phthalate (DEHP), di-isobutyl phthalate (DIBP), and diallyl phthalate (DAP) are or have been used as well. Phthalates undergo rapid metabolism and in addition to forming specific metabolites share phthalic acid (PA) as a common metabolite (Albro et al. 1984). In several studies phthalate monoesters metabolites have been used as markers of exposure (Blount et al. 2000; Silva et al. 2004). However, for several high molecular weight phthalates (for instance DEHP and DOP) no specific metabolites have been unequivocally identified (Kato et al. 2005). Until specific biomarkers of exposure to isomeric phthalates are available, indirect measures of exposure (e.g. phthalic acid) to these phthalates might be valuable. Furthermore, in a recent study in the Swedish rubber industry significant correlations were found between free urinary PA, on the one hand, and mono-ethyl phthalate, mono- *n*-butyl phthalate, mono-benzyl phthalate, and mono ethylhexyl phthalate, on the other (Bo A.G. Jönsson, personal communication). Similarly, in a study among anonymous volunteers a strong correlation ($r = 0.85$) was observed between the concentration of total PA and the sum of the concentration of 13 phthalate metabolites (Albro et al. 1984). From these studies it can be concluded that indeed hydrolysed or non-hydrolysed PA can be used as an indicator of total phthalate body burden.

Historically, carbon disulphide (CS_2) has been used as a solvent in the rubber industry. However, nowadays CS_2 exposure in the rubber industry mainly arises from the formation of CS_2 during curing due to decomposition of dithiocarbamates and thiurams, which are used as accelerators. In addition, exposure to dithiocarbamates itself has been shown to produce CS_2 in vivo (Johnson et al. 1996). After exposure to CS_2, 2-thiothiazolidine-4-carboxyl acid (TTCA) is generated in the body via conjugation with glutathione (Graham et al. 1995). Given the considerable specificity and sensitivity of urinary TTCA for CS_2 exposure, and overall low background level in the non-exposed population, it has been generally utilized as a biomarker of exposure to CS_2. Biomonitoring action levels (BALs) for TTCA have been given in many countries (Kivisto 2000). The American Conference of Governmental Industrial Hygienist (ACGIH) in the US adopted 5 mg/g creatinine (Cr.) (3500 μmol/mol Cr.) in the urine as the biological exposure index (BEI) for urine samples collected at the end of the work shift, corresponding with a time-weighted exposure level of 10 ppm (ACGIH 2002)

The present study was designed to explore urinary levels of TTCA and PA across factories and departments in the contemporary rubber manufacturing industry. Besides providing information on contemporary exposures to CS_2 and total phthalates the results should offer insight in how phthalate or CS_2 exposure could be assessed in an epidemiological study. For this purpose 386 urine samples from 101 rubber workers were selected from a large cross-sectional exposure survey conducted in the Netherlands (Vermeulen et al. 2000a).

Materials and methods

Study population

Selection of the study population is described in detail elsewhere (Vermeulen et al. 2000b). In short, 224 male subjects, employed in nine rubber manufacturing companies (three rubber tire, five general rubber goods, and one retreading company), were randomly selected based on their department affiliation (e.g., compounding and mixing, pretreating, molding, curing, finishing, shipping, and engineering service). This selection procedure ensured coverage of all available departments within the study companies. Sample and data collection was conducted from January to July 1997 and was carried out within one week in each company. It comprised a medical survey, area and personal exposure measurements, and collection of biological samples. Participation was voluntary, and written informed consent was obtained after the study was explained to the subjects. For the purpose of this study an approximate 50% stratified random sample from the initial population was drawn that covered all factories and departments (Vermeulen et al. 2001)

Urine collection

Spot urine samples were collected on Sunday, Tuesday, Wednesday, and Thursday at approximately the same time of the day (±4 pm). Samples were stored at −20°C till analyses (~6 years). The Cr. levels were measured using the Jaffe reaction with a Hitachi autoanalyzer.

Analysis of phthalic acid and thiothiazolidine-2-carboxylic acid

PA and TTCA were extracted from urine by the same method. Aliquots of 500 ng deuterium labeled PA (Aldrich, Milwaukee, WI, USA) in 0.5% acetic acid was added to 1 ml of urine as an internal standard. Calibration standards were prepared by spiking urine from unexposed subjects with know amounts of PA (Merck, Darmstadt, Germany) and TTCA (Aldrich). The urine samples were acidified with 0.1 ml 6 M hydrochloric acid and the ion strength raised with 0.5 ml 6 M sodium chloride. The samples were shaken with 6 ml ethyl acetate, the organic phase separated, evaporated and the samples re-dissolved in 0.5 ml water containing 0.5% acetic acid. Analyses were performed with a Perkin Elmer Series 200 liquid chromatography system with autosampler (Applied Biosystems, Norfolk, CT, USA), coupled to an API 3000 triple quadrupole mass spectrometer (Applied Biosystems/MDS-SCIEX, Toronto, Canada) with a Turbo electrospray ion source in negative ion mode at 350°C. The column was a Genesis C_{18}(50 × 2.1 mm) with a particle size of 4 μm (Jones, Lakewood, CO, USA). The mobile phases were (A) water containing 0.5% acetic acid and (B) methanol containing 0.5% acetic acid.

For PA an isocratic elution with 30% B during 3.5 min followed by a gradient up to 100% B during 2 min was used. The fragments analyzed were for PA m/z 164.7/76.9 at a collision energy (CE) at −23 V and for deuterium labeled PA m/z 168.7/81.0 (CE −25 V). The declustering potential was −26 V. The limit of detection (LOD) was 5 ng/ml and the precision, determined from duplicate analysis of the same samples on different days, were 9% at 50 ng/ml and 10% at 300 ng/ml.

For TTCA an isocratic elution with 5% B during 3 min followed by a gradient up to 100% B during 2 min was used. The fragments analyzed were m/z 161.9/57.9 at a CE at −14 V and a declustering potential at −26 V. The LOD was 1 ng/ml and the precision, determined from duplicate analysis of the same samples on different days, were 11% at 10 ng/ml and 7% at 70 ng/ml.

Technicians involved in the analyses were not aware of the identity of any samples or related conditions. Urine from normal subjects was used as quality control and analyzed together with the samples. The analyses of TTCA were part of the Round Robin intercomparison program (Professor Dr. med. Hans Drexler, Institute and Out-Patient Clinic for Occupational, Social and Environmental Medicine, University of Erlangen-Nuremberg) with analysis results within the tolerance limits. No such program exists for PA to our knowledge.

Non-detectable urinary levels of PA (2 out of 386) and TTCA (26 out of 386) were replaced with values equal to the LOD divided by the square root of 2 (Hornung and Reed 1990).

Personal exposure measures

Personal inhalable particulate exposure was measured on the days of urine collection (e.g. Tuesday, Wednesday, and Thursday) by means of a PAS6 sampler (Kenny et al. 1997). Personal dermal exposure to cyclohexane soluble matter (CSM) was measured with a dermal pad sampler worn at the lower part of the wrist of the hand of preference (Vermeulen et al. 2000a). The CSM contents of inhalable (e.g. rubber fumes) and dermal exposure samples were determined by means of the NIOSH PCAM 217 method (NIOSH 1977). Less than 5% of the subjects used any kind of respiratory protection during the measurements. Gloves were used by approximately 45% of the study population.

Statistical analyses

Urinary levels of PA (μg/l) and TTCA (μmol/mol creatinine) are summarized as averages (AM), geometric means (GM), and geometric standard deviations (GSD). Normal probability plots indicated that urinary levels of PA and TTCA could be best described by lognormal distributions. Therefore, natural logarithms of exposure concentrations were used in the statistical analyses. Correlations between the two urinary biomarkers and between biomarker levels by day of urine collection were calculated using the Pearson correlation statistic. Differences in biomarker levels between sampling days and between Sunday urinary biomarker levels and the mean individual's weekday urinary biomarker level were tested for statistical significance by paired t-test.

To further study the effect of factory, department, personal exposure measures (e.g. inhalable dust, rubber fumes, and dermal exposure) and lifestyle factors we used multivariate mixed-effect models. In these models 'factory', 'department', the natural logarithm of personal exposure measurement and/or lifestyle factors (e.g. age and smoking habits) were included as *fixed effects* while 'worker' was included as *random effect* to account for repeated measures on the same person. These models were corrected for individual biomarker background levels by adding the Sunday urinary biomarker as an additional *fixed effect* in the model. Model fit was evaluated using the Akaike Information Criteria (AIC).

All analyses were carried out using SAS Version 8.0 software (SAS Institute, Cary, NC, USA). Statistical significance was set at $P < 0.05$.

Results

Of the selected rubber workers 101 subjects (87%) had a valid Sunday measure of urinary PA and TTCA. Results presented in this paper are therefore limited to this population. In total 386 urine samples were successfully collected and analyzed.

Urinary levels of PA and TTCA increased significantly during the workweek compared to Sunday (Table 1). For both biomarkers the average weekday levels were about a factor 2 higher than the Sunday urinary levels (based on the geometric means). No statistical differences in biomarker levels could be detected between the different days of the workweek except for urinary TTCA levels on Wednesday and Thursday (Paired t-test, $P = 0.037$ ($k = 91$)). For the sub-population ($k = 85$, $n = 340$) that had valid measures of both biomarkers on all days (e.g. Sunday, Tuesday, Wednesday, and Thursday), patterns were essentially identical to the overall population with increased levels during the workweek as compared to the Sunday values and little to no differences between workdays. However, it is clear that significant background levels of in particular PA and to a lesser extent TTCA were present in samples collected on Sunday, which were collected approximately 48 h after exposure had ended. Sunday urinary PA levels correlated weakly with weekday samples ($r = 0.22$–0.31) (Table 2). Not surprisingly, levels on the different weekdays correlated strongly ($r = 0.87$–0.90). The picture for TTCA was slightly different with no significant correlation between Sunday and

Table 1 Urinary levels of phthalic acid (*PA*) and 2-Thiothiazolidine-4-carboxylic acid (*TTCA*) in rubber workers by day of urine collection

	N	AM	GM	GSD	Minimum	Maximum	*P*-value[a] (*t*-test)
PA (µg/l)							
Sunday	101	136	83	2.49	4	2449	
Tuesday	95	303	148	3.16	7	4894	< 0.0001
Wednesday	93	352	152	2.95	25	9533	< 0.0001
Thursday	97	348	164	2.73	24	10140	< 0.0001
TTCA (µmol/mol Cr.)							
Sunday	101	18	6	4.61	0.3	221	
Tuesday	95	22	11	3.77	0.2	192	0.0004
Wednesday	93	32	13	4.00	0.2	780	0.0002
Thursday	97	23	9	4.57	0.2	381	0.0454

[a] Tested against the Sunday biomarker level (Paired *t*-test)

Table 2 Pearson correlation between urinary levels of phthalic acid (*PA*) and 2-Thiothiazolidine 4-carboxylic acid (*TTCA*) in rubber workers by day of urine collection (*P*-value between parenthesis)

	Tuesday	Wednesday	Thursday
PA (µg/l)			
Sunday	0.31 (0.0003) $n = 95$	0.30 (0.0033) $n = 93$	0.22 (0.030) $n = 97$
Tuesday		0.90 (< 0.0001) $n = 87$	0.87 (< 0.0001) $n = 91$
Wednesday			0.90 (< 0.0001) $n = 91$
TTCA (µmol/mol Cr.)			
Sunday	0.17 (0.10) $n = 95$	0.08 (0.42) $n = 93$	0.06 (0.59) $n = 97$
Tuesday		0.43 (< 0.0001) $n = 87$	0.53 (< 0.0001) $n = 91$
Wednesday			0.45 (< 0.0001) $n = 91$

weekday urinary levels ($r = 0.06$–0.17) and moderate correlation between the different days of the week ($r = 0.43$–0.53). Noteworthy, no correlation between the two biomarkers was observed during the workweek ($r = 0.12$).

Mean weekday urinary biomarker levels by factory and department are presented in Table 3. Levels of PA were generally elevated compared to the background level (83 µg/l). However, the increase in PA levels during the week as compared to Sunday was not uniform across factories and departments and only reached statistical significance for factory D and H and for the departments 'curing' and 'molding'. Urinary PA levels for subject in 'pre-treating' seemed also elevated but formal statistical significance was not reached. Similarly, TTCA levels were generally elevated compared to the average background level (6 µmol/mol Cr.) but reached statistical significance only for factory C and D and the departments 'curing' and 'molding'.

The association between factories and departments and urinary biomarker levels was further explored in multivariate models. As no association was found between lifestyle factors (e.g. current smoking habits and age) or personal protective equipment (e.g. respiratory or dermal protection) and urinary PA or TTCA, the presented models are not corrected for these factors. Including factory in the mixed model resulted in a better model fit for both markers than department (Table 4). Combining both factory and department in the model as main effects showed that both were predictors for urinary PA but that no significant association was found between departments and urinary TTCA levels indicating that the elevated TTCA levels were more determined by factory than by department (e.g. curing and molding). No statistical significant interaction was found between department and factory.

As exposures were highest in the curing department we explored if the biomarker levels had any relation to exposure measures that had been collected more routinely in the rubber industry (Kromhout et al. 1994; Vermeulen et al. 2000a). No association was found between inhalable dust, rubber fumes measured as CSM, dermal CSM and any of the urinary markers (Table 5).

Discussion

We studied the range in urinary levels of TTCA and PA across factories and departments in the contemporary rubber manufacturing industry. For this purpose 386 urine samples from 101 rubber workers were analyzed. Urinary concentrations of TTCA were adjusted for Cr. contents. However, results based on volume adjusted biomarker concentrations rendered essentially the same results (Data not shown).

In our study design, Sunday urinary values were used as individual background (e.g. reference) levels. Given the relatively short half-life of both TTCA (~8 h) and PA (~5 h) there should be minimal contribution of the preceding Friday workplace exposures to the Sunday

Table 3 Mean weekday urinary levels of phthalic acid (*PA*) and 2-Thiothiazolidine-4-carboxylic acid (*TTCA*) in rubber workers by factory and department

	K^a	N^b	PA (µg/l)			TTCA (µmol/mol Cr.)		
			AM	GM	GSD	AM	GM	GSD
Factory								
A	8	23	89	65	2.39	49	17	5.94
B	12	36	253	168	2.46	14	8	3.61
C	7	20	271	139	2.61	35	26***	3.12
D	9	26	1537	719**	3.09	73	30**	3.20
E	13	38	163	114	2.39	22	10	5.20
F	15	41	154	97	2.64	21	11	3.08
G	8	22	147	123	1.78	15	9	3.06
H	17	49	379	272****	2.25	10	7	2.84
I	12	30	149	109	2.25	17	7	4.80
Department								
Mixing	10	30	170	137	1.90	15	7	4.43
Pre-treating	14	41	341	163	3.00	16	8	3.86
Molding	27	76	199	111**	2.82	34	11*	4.16
Curing	24	67	742	288***	3.83	27	16*	3.11
Finishing	9	25	186	140	2.29	42	13	5.80
Shipping	3	8	107	92	1.86	15	14	1.90
Eng. Service	14	38	157	105	2.57	17	7	4.67

Mean weekday urinary biomarker levels of subjects in a particular factory or department significantly higher than Sunday urinary biomarker levels (paired *t*-test): * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; **** $p < 0.0001$

[a] Total number of subjects ($k = 101$)

[b] Total number of weekday measurements ($n = 285$)

afternoon sample under one compartment linear kinetics, 1.6 and 0.1%, respectively. Nevertheless, PA was detected in 98% of the Sunday urine samples (> 5 ng/ml), while in 87% detectable levels (> 1 ng/ml) of TTCA were measured. The high background levels for PA are unlikely to be due to contamination of collection bottles, laboratory supplies and equipment as the total contribution of all these factors was recently estimated to be at most around 2 µg/l total PA (Kato et al. 2005), which is considerably lower than the levels measured in this study and is similar to the background levels found in our study. These results thus indicate that environmental and/or lifestyle factors contribute to both urinary biomarker levels. Phthalates are ubiquitous environmental chemicals and as such the high frequency of detectable levels is not surprising. Hence, more than 75% of the general US population screened in the NHANES survey had detectable levels of specific phthalate metabolites (Silva et al. 2004). It has been hypothesized that despite the rapid metabolism and elimination of most phthalates, a very stable background concentration may in theory be reached through chronic low-level exposures from dietary ingestion and many commonly used products (Duty et al. 2003; Silva et al. 2004). This is to some extend corroborated in our study as we reported moderate but significant correlations between Sunday and weekday urinary PA concentrations indicating the presence of relatively stable individual background concentration in urinary PA concentrations. For TTCA such an association was not found. Background TTCA concentrations have been associated with dietary factors with most noticeably high concentrations of endogenous TTCA levels in cruciferous vegetables, which is excreted

Table 4 *P*-values for type 3 tests of fixed effects of factory and department on phthalic acid (*PA*) and 2-Thiothiazolidine-4-carboxylic acid (*TTCA*) based on mixed effect models correcting for individual Sunday urinary biomarker values ($k = 101$, $n = 285$)

Model	Fixed effect	PA ln(µg/l) (*P*-values)	TTCA ln (µmol/mol Cr.) (*P*-values)
1	Factory	< 0.0001	< 0.0001
2	Department	< 0.0001	< 0.0001
3	Factory	< 0.0001	0.011
	Department	0.0020	0.28
4	Factory	< 0.0001	0.063
	Department	0.0026	0.36
	Factory × Department	0.26	0.14

unchanged in the urine (Kivisto 2000). Background levels on a given day will therefore depend on the precise diet during the day.

The PA and TTCA increased during the workweek relative to the individual Sunday biomarker values indicating that workers in the contemporary rubber industry are exposed to phthalates and CS_2. Based on linear assumptions between CS_2 exposure and urinary TTCA it can be estimated that exposure levels of CS_2 on average were below 0.05 ppm. Noteworthy, none of the urinary TTCA concentrations in our study exceeded the BEI of 3500 µmol/mol Cr. (Max 780 µmol/mol Cr.). The results of this study are in agreement with an unpublished study in a Swedish rubber plant performed in 2002 (Bo A.G. Jönsson, personal communication). In this study, the median urinary TTCA levels among unexposed subjects ($k = 120$) was 5 µmol/mol Cr. compared to 6 µmol/mol Cr. (~8 µg/l) in the 'unex-

Table 5 Association between personal inhalable dust, rubber fumes measured as cyclohexane soluble matter (*CSM*), dermal CSM exposure and urinary levels of phthalic acid (*PA*) and 2-Thiothiazolidine-4-carboxylic acid (*TTCA*) among curing workers ($k = 24$)

Exposure[a]	N	PA (μg/l)		TTCA (μmol/mol Cr.)	
		β (SE)	*P*-value	β (SE)	*P*-value
Inhalable dust (mg/m^3)	62	0.11 (0.21)	0.58	−0.061 (0.38)	0.44
Rubber fumes (CSM μg/m^3)	62	0.078 (0.12)	0.52	−0.064 (0.25)	0.78
Dermal CSM	66	0.0064(0.072)	0.38	0.015 (0.14)	0.92

[a] Exposures were entered in separate models as continues variables after natural log transformation

posed' Sunday samples in our study. Median TTCA levels among the Swedish rubber workers ($k = 180$) was 25 μmol/mol Cr., which is equivalent to levels detected in urine of rubber workers in our higher exposed factories (Median TTCA among all subjects was 13 μmol/mol Cr. (~29 μg/l)). Median PA concentrationf in the Swedish study among the controls was 50 μmol/mol Cr., which is almost identical to the median level in the Sunday samples (median: 77 μg/l ~47 μmol/mol Cr.) in our study. Interestingly, no significant difference in urinary PA was found in the Swedish study between the exposed workers and the controls. The almost identical background levels of both markers between the two studies corroborate however, that the Sunday urinary values could indeed be regarded as unexposed samples and that most likely urinary markers had remained stable during the several years of storage as, in contrast to our study, the Swedish samples were analyzed shortly after collection.

Multivariate analyses confirmed that differences in phthalate and CS_2 exposure were mostly driven by factory and to some extent by departments as well. The factories with the highest exposure to phthalates, as measured by PA, were a general rubber goods plant producing mainly rubber to metal bonded products and a tire factory. No detailed information was available on which phthalate containing ester plasticizers were actually used at the time of measurement in these two plants due to trade secrecies. Analyses of specific phthalate metabolites should however shed further light on the speciation of phthalate monomers as toxicological properties of phthalates are highly variable. Exposure to phthalates within factories seemed to increase with increasing production temperatures, which increases during pre-treating, molding and are the highest in curing, due to off gassing of phthalates. The same pattern was observed for TTCA with the highest exposures in the curing department. In this case the highest exposures were observed in two general rubber good factories that had a large number of small compression presses. The CS_2 sources in this case were most likely several different dithiocarbamate (TDEC, ZDMC, ZDEC, and ZDBC) and thiuram (TMTD, TETD) compounds, which release CS_2 during curing. Some CS_2 might have been formed in vivo after dithiocarbamate exposure (Johnson et al. 1996). However, this seems unlikely as exposure to dithiocarbamates would have been highest during weighing and mixing of the raw chemicals and no increase in TTCA was observed within this department. As such the increased levels in this study can be most likely directly related to airborne CS_2 exposure.

The results of our study show that rubber workers in the contemporary rubber industry are exposed to low levels of phthalates and CS_2 (~0.05 ppm), as measured by urinary PA and TTCA. Exposures to both compounds are largely driven by specific circumstances in factories. Therefore, if estimating exposures for epidemiological studies, detailed information should be collected on the type and amount of phthalates, dithiocarbamates and thiurams used. This might provide serious limitations as companies are not often forthcoming about their exact composition of rubber materials (as in this study), the composition might change overtime and employees might have difficulties identifying, which chemicals and in what quantities they were used, especially if they were not involved in the mixing and compounding. Furthermore, no correlations were found with more routinely collected exposure measures in the rubber industry (e.g. rubber dust and fumes) and as such these cannot be used as valid surrogates. It seems therefore that in order to study the effect of phthalates and CS_2 in the contemporary rubber industry exposure measurements should be collected in prospective or cross-sectional studies. In this case, biological monitoring seems a reasonable approach. However, attention should be given to the individual background levels of these markers as they could lead to a substantial overestimation of the occupational contribution to the total body burden of CS_2 but especially phthalate exposure.

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